Cucumber Pollen Germination and Tube Elongation Inhibited or Reduced by Pesticides and Adjuvants[1]

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ABSTRACT

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Commercial formulations of 5 insecticides: diazinon, dicofol, endosulfan, malathion, and Pyrenone* (6% pyrethrins and 60% piperomyl butoxide); 2 fungicides: captan and Manzate* (zinc and manganese ethylene bisdithiocarbamate); and 9 adjuvants: Fomex*, Multi-Film X-77*, Nu-Trex*, Dupont Spreader Sticker*, Bio-88*, Target-E*, Bio-Film*, Dupont Surfactant F*, and Regulaid* severely reduced germination of cucumber pollen on an artificial medium. A lesser degree of reduction in germination was caused by the insecticides carbaryl and methoxychlor, the fungicide benomyl, and the adjuvants Regulaid*, Triton B-1956* and Chevron Spreader Sticker*. Carbaryl, methoxychlor and benomyl were used at spray concentrations of active ingredients recommended on the labels in 100 gal (378.5 liters) of water. The remainder of the pesticides was used at concentrations below label recommendations.

Diazinon applied under controlled conditions to hand-pollinated pistillate flowers of cucumbers in greenhouse caused parthenocarpy and fruit abortion.

Results of investigations on the effect of various pesticides and adjuvants on pollen of plants in vivo and in vitro have been reported by several investigators up to 1972, and were reviewed by the present investigator (Gentile et al. 1971, Gentile and Gallagher 1972, Gentile et al. 1972). Additional investigations have been reviewed more recently by McGregor, 1976, together with a review of the pollination requirements of cucumber and other plants.

The present investigations were prompted by reports of poor fruit set in cucumber fields in 1973–74 in Massachusetts. Tentatively this was ascribed to poor pollination resulting from the killing of bees and other pollinators by pesticides. The possibility of an adverse effect of pesticides on pollen germination was also suggested. In the absence of any published material on this subject in relation to cucumbers, the present in vivo and in vitro investigations under greenhouse and laboratory conditions were conducted at the Suburban Experiment Station of the University of Massachusetts at Waltham.

In the in vitro evaluation, the following insecticides, fungicides, and adjuvants were used. Insecticides: Carbaryl 50% WP, Diazinon 50% WP, Dicofol 35% WP, Endosulfan 24% WP, Malathion 25% WP, Methoxychlor 50% WP, and Pyrenone* EC (pyrethrins 6%, piperonyl butoxide 60%). Fungicides: Benomyl 50% WP, Captan 50% WP, and Manzate 200 80% WP (zinc and manganese ethylene bisdithiocarbamate). Adjuvants: Bio-88*—alkylpolyethoxyethanol, free fatty acids, isopropanol; Bio-film*—alkylarylpolyethoxyethanol, free and combined fatty acids, glycol ethers, di-alkyl benzenedicarboxylate, isopropanol; Fomex*—alcohol sulfate, salts of alkyl and di-alkyl2, 5-diketotetra- hydrofuran, alkyl sulfonates, isopropanol; Multi-Film* X 77—alkylarylpolyoxyethylene glycols, free fatty acids, isopropanol; Nu-Trex*—alcohol ethoxylates, polyhydric alcohols, glycol ethers, phosphoric acid, sulfates of zinc, iron, copper, and manganese; Regulaid*—polyoxyethylenepolypropoxypropanol, alkyl 2-ethoxyethanol; Chevron Spray-Sticker*—alkyl olefin aromatic polymers; Dupont Spreader-Sticker*—sodium sulfates of mixed long chain alcohol fatty acid esters, diethylene glycol abietate; Dupont Surfactant F*—details on formulation not provided by manufacturer; Target E*—blend of industrial invert disaccharides solubilizers, stabilizers, non-ionic emulsifier plus preservative in a water base solution; and Triton B 1956*—modified phtalic glycerol alkid resin.

Most of the compounds were selected because of their actual or potential utilization in cucumber pest control programs. The adjuvants were used at the highest rate of formulation suggested by manufacturers for general field applications in 100 gal (378.5 liters) of water (see Table 3). The fungicides were used at the following rates in ppm AI in 100 gal (378.5 liters) of water: captan 50% WP, 1198 ppm or 2 lb (907.2 g) formulation; Manzate 200 80% WP, 1916.8 ppm or 2 lb (907.2 g) formulation; benomyl 50% WP, 224.7 ppm or 6 oz (170.1 g) formulation. On a per acre (0.405 ha) basis, only captan and Manzate were used at rates below label recommendations. The insecticides were used at the rate of 1000 ppm AI. With the exception of carbaryl and methoxychlor, this approximates half of the respective rates suggested on the label for field application in 100 gal (378.5 liters) of water. Diazinon, the only pesticide used in the in vivo evaluations, was also used at the 1000 ppm rate.

The cucumber cv. 'Marketmore 70' was used as a source of pollen and pistillate flowers in the in vitro and in vivo greenhouse experiments with insecticides. The field grown cultivar Spartan Salad was used as a source of pollen in the in vitro experiments with the surfactants and fungicides. For the in vitro experiments, newly-opened staminate flowers were gathered during the early morning hours into plastic bags and refrigerated for 2–4 h. The pollen germination medium, modified after Brewbaker and Kewack (1963), consisted of 92% sucrose, 7.35% agar, 0.28% calcium nitrate, 0.18% magnesium sulfate, 0.09% boric acid, and 0.09% potassium nitrate in distilled water. The medium solution was heated to near boiling point, then poured into sterile Petri dishes to ½ their depth and allowed to solidify. (The medium may be refrigerated for 2–3 wk). Discs of solidified medium were used in the pollen germination tests. Up to 5 discs were transferred with a No. 9 cork borer and a spatula onto a microscope slide overlaying a moistened filter paper in a sterile Petri dish. The upper surface

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* Trade name.

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of each disc was treated topically with ½ ml of solution or suspension of pesticide or adjuvant and the excess water allowed to evaporate at room temperature. The control discs received distilled water. The cucumber pollen was transferred from flowers with newly dehiscing anthers onto the treated discs of medium with a camel's hair brush. The dishes were closed and kept at room temperature for 1–2 h, followed by refrigeration until the germination counts were made. The percentage of pollen germination was determined from random counts of 100 pollen grains per each of 5 discs made with a compound scope. The avg length of the pollen tubes was derived from 10 measurements per each of 5 discs at 150 X. Each evaluation was repeated 3 times.

In the in vivo experiments conducted in late fall and winter under greenhouse conditions at a temperature of ca. 21°C, the cucumber plants were grown in plastic pots and watered and fed as needed. Pistillate buds just prior to flowering were capped, usually in late afternoon, with gelatin capsules (Fig. 1) and tagged according to the following categories: (a) hand pollinated diazinon-treated; (b) hand pollinated water-treated control; and (c) not pollinated and untreated control. The pistillate flowers of the 1st 2 categories were uncapped and treated with diazinon or distilled water with an atomizer. These treated pistillate flowers were pollinated with pollen from newly dehiscing anthers and then recapped. The pollen used in the experiment had yielded a 93% germination in vitro. Pollination began in late Oct. and was terminated in Nov. Pistillate flowers were assigned to the 3 categories on each tagging day to assure chronological representation. Twenty–25 pistillate flowers of each category were tagged. The tags were removed from the treated and pollinated flowers, and from the untreated and not pollinated control flowers when the pistils seemed no longer receptive to pollination.

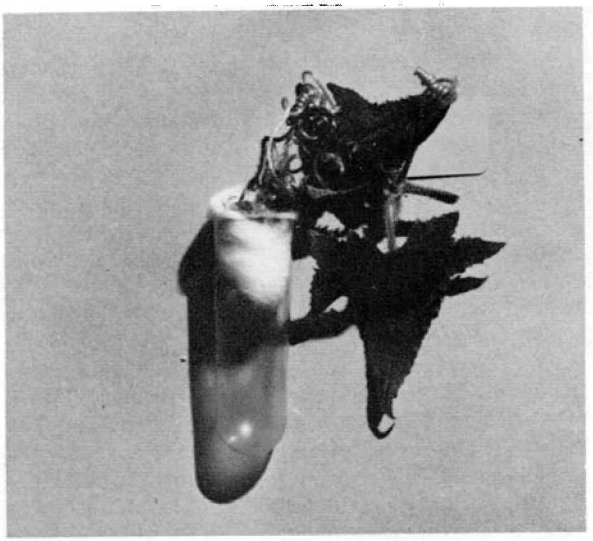

FIG. 1.—Method of capping pistillate flowers.

In the evaluation of the in vivo experiment, the fruits that had set and developed normally were harvested at maturity and checked for normal seed production. Abnormal fruits, mostly characterized by proximal and distal tapering, or by small size, were checked for parthenocarpy. Flowers that failed to set fruits were categorized as aborted.

Results and Discussion

The data summarized in Table 1 indicate that the dry residue of commercial formulations of dicofol WP, diazinon WP, malathion WP, endosulfan EC, and Pyrenone EC in an increasing order of severity inhibit germination and pollen tube elongation in cucumber when topically applied to agar medium at 1000 ppm AI concn in distilled water. In the case of Pyrenone, the medium was treated with a 1000 ppm concn of pyrethrins and 10,000 ppm piperonyl butoxide.

A lesser degree of inhibition of germination was observed with 1000 ppm concn of carbaryl WP and methoxychlor WP. It is worth mentioning that, with the exception of carbaryl and methoxychlor, the above chemicals were used at half the rate suggested on the label for field application in 100 gal of water.

Table 1.—Germination and tube elongation of cucumber pollen on artificial medium topically treated with 1000 ppm AI of commercial insecticides.

Treatment	% germination 1500 grains/ treatment	Avg length of pollen tube in mm 150 grains/ treatment
Dicofol 35% WP	20.0	0.020
Diazinon 50% WP	25.3	.016
Malathion 25% WP	31.1	.015
Endosulfan 24% EC	49.5	.015
Pyrenone (pyrethrins 6%, tech. piperonyl butoxide 60%)[a]	59.6	.010
Carbaryl 50% WP	65.33	.131
Methoxychlor 50% WP	70.6	.118
Control (distilled water)	90.7	.185

[a] Medium received 1000 ppm pyrethrins and 10,000 ppm piperonyl butoxide.

Table 2 summarizes the results obtained with commercial WP formulations of 3 fungicides. The data indicate that captan and Manzate, at rates below label recommendation, severely inhibited pollen germination and tube elongation in vitro, whereas benomyl used at a rate recommended on the label caused a lesser degree of inhibition.

Table 2.—Germination and tube elongation of cucumber pollen on artificial medium topically treated with 3 commercial fungicides.

Treatment and ppm AI	% germination 2500 grains/treatment	Avg length of tube in mm No. grains in parentheses
Captan 50% WP 1198 ppm	7.2	0.19 (121)
Manzate 80% WP 1916 ppm	8.01	.11 (94)
Benomyl 50% WP 224.7 ppm	61.84	.19 (250)
Control (distilled water)	83.4	.35 (250)

Table 3 summarizes the results obtained in vitro with commercial formulations of adjuants used at rates suggested on the labels in 100 gal (378.5 liters) of water. The data indicate that Fomex, Multi-Film X77, Nu-Trex, Dupont Spreader-Sticker, and Bio-88 severely inhibited germination and pollen tube elongation, whereas Target-E, Bio-

Table 3.—Germination and tube elongation of cucumber pollen on artificial medium topically treated with label-suggested concentrations of pesticide adjuvants.

Treatment	Amt. of formulation in fl oz/100 gal of water		% germination 500 grains/ treatment	Avg length of tube in mm 50 grains/treatment
Fomex	96	(2.83)[b]	9.2	0.33
Multi-Film X77	8	(0.237)	12.7	.13
Nu-Trex	32	(0.946)	15.7	.19
Dupont Spreader-Sticker	4	(0.118)	16.8	.22
Bio-88	8	(0.237)	17.7	.13
Target E	320	(9.47)	51.6	.08
Bio-Film	8	(0.237)	54.1	.16
Dupont Surfactant F	4	(0.118)	56.0	.16
Regulaid	48	(1.42)	61.7	.16
Triton B-1956	5	(0.148)	70.4	.18
Chevron Spreader-Sticker	8	(0.237)	73.6	.19
Control	Distilled Water		78.7[a]	.23[a]

[a]Avg of 3 tests.
[b]Liters of formulation in 378.5 liters of water.

Film, and Dupont Surfactant F caused a lesser degree of inhibition. The adjuvants Regulaid, Triton B-1956 and Chevron Spreader-Sticker were the least deleterious to the cucumber pollen.

Table 4 summarizes and Fig. 2 shows the results of a greenhouse test aimed at corroborating the results obtained in vitro with diazinon. The procedures used in this study proved to be quite laborious and, consequently, the number of replications was kept to a minimum. The data gathered indicate that hand pollination of pistillate flowers previously treated with 1000 ppm concn of diazinon gave rise to a high incidence of aborted and parthenocarpic fruits similar to that observed in the untreated and not pollinated control.

Table 4.—Parthenocarpy or fruit abortion caused by 1000 ppm Diazinon applied to cucumber flowers prior to hand pollination.[a]

Treatment	Aborted and parthenocarpic fruits	Fruits with normal seeds
Diazinon treated flowers, hand pollinated	24	1
Water treated flowers, hand pollinated	13	12
Untreated flowers, not hand pollinated	20	—

[a]Percent germination of pollen in vitro: 93.0.

Several of the pesticides and adjuvants causing adverse effects on cucumber pollen in this study had proved in previous studies to be similarly deleterious to the pollen of petunia, tomato, and sweet corn; however, it is worth noting that in previous studies 1000 ppm concn of carbaryl caused severe inhibition of germination in sweet corn, and that benomyl at 1000 ppm concn severely inhibited germination of petunia pollen. It should be mentioned that, among the adjuvants Triton B-1956 and Regulaid in previous tests caused the least inhibition of pollen germination.

It is hoped that these observations will stimulate investigations on the number of viable pollen grains and relative tube length needed to assure normal fruit and seed set in cucumber, and on the relevance of the role of pesticides in interfering with the proper completion of these processes under greenhouse and field conditions. The action of chemicals in improving yield and fruit set by delaying fertilization of inflorescences should also be investigated.

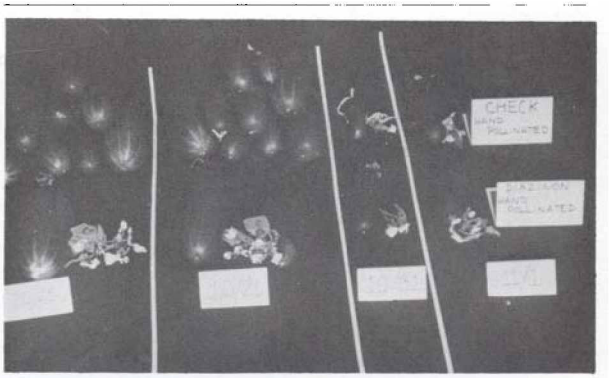

FIG. 2.—Top row; fruits from water-treated check; 2nd row; fruits from diazinon-treated pistillate flowers. Dates of hand-pollination shown below 2nd row.

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